

3 a plurality of fiber optic cables for illuminating volumes of the plurality of samples,
4 a plurality of lenses, each co-axially disposed with a first end of a fiber optic cable for
5 focusing an excitation beam into a sample, and
6 a fiber optic multiplexer which couples the detection and analysis mechanism to a
7 second end of each of the plurality of fiber optic cables.

1 16.⁴ The apparatus according to claim 13¹ wherein the sample holder includes a
2 removable reaction chamber for holding the sample.

1 17. ~~The apparatus according to claim 16 wherein the removable reaction chamber is~~
2 ~~sealable.~~

1 18.⁶ The apparatus according to claim 13¹ wherein the sample holder includes a sealable
2 reaction chamber for holding the sample.

1 19.⁷ The apparatus according to claim 13¹ wherein the sample holder includes an optical
2 interface through which the excitation beam is transmitted from the lens into the sample.

1 20.⁸ The apparatus according to claim 19⁷ wherein the sample holder includes a sealable
2 reaction chamber for holding the sample, the optical interface forming a wall of the reaction
3 chamber.

1 21.⁹ The apparatus according to claim 19⁷ wherein the apparatus further includes a
2 mechanism for heating the optical interface to prevent condensation of the sample on the
3 optical interface.

1 22.¹⁰ The apparatus according to claim 21⁹ wherein the sample holder includes a sealable
2 reaction chamber for holding the sample, the optical interface forming a wall of the reaction
3 chamber.

1 23.¹¹ The apparatus according to claim 19⁷ wherein the sample holder includes a

2 removable reaction chamber for holding the sample, the optical interface forming a wall of
3 the reaction chamber which covers at least a portion of the sample and which is separated
4 from the sample by an air gap.

1 24. A method for monitoring the formation of a nucleic acid amplification reaction product
2 in real time comprising:

3 adding a sample to a sample holder which contains a nucleic acid sequence to be
4 amplified,

5 transmitting an excitation beam into the sample which illuminates a volume of the
6 sample, the sample including a first fluorescent indicator which produces a first fluorescent
7 signal when illuminated by the excitation beam whose intensity is proportional to the
8 concentration of amplification reaction product in the sample and the volume of the sample
9 illuminated by the excitation beam, and a second fluorescent indicator homogeneously
10 distributed throughout the sample which produces a second fluorescent signal when
11 illuminated by the excitation beam whose intensity is proportional to the volume of the
12 sample illuminated by the excitation beam; and

13 measuring the intensities of the first and second fluorescent signals.

1 25. The method according to claim 24 wherein the first and second fluorescent signals
2 each have an intensity and the detection, the step of measuring the intensities of the first
3 and second fluorescent signals including calculating a ratio between the intensity of the first
4 fluorescent signal and the intensity of the second fluorescent signal.

1 26. The method according to claim 24 wherein the first fluorescent indicator is a
2 complex-forming dye.

1 27. The method according to claim 24, further including the step of sealing the sample
2 within the sample holder prior to transmitting an excitation beam into the sample.

1 28. The method according to claim 24 wherein the sample holder includes an optical
2 interface through which the excitation beam is transmitted from the lens to the sample, the

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3 sample holder also including an air gap separating the optical interface from the sample, the
4 method further including the step of heating the optical interface to prevent condensation of
5 the sample on the optical interface.

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1 29.16 The method according to claim 28, further including the step of sealing the sample
2 within the sample holder prior to transmitting an excitation beam into the sample.

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1 30. The method according to claim 24 wherein the step of adding a sample to a sample
2 holder includes
3 adding a sample to a reaction chamber which is removable from the sample holder;
4 and
5 adding the removable reaction chamber to the sample holder.

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1 31.18 The method according to claim 30, further including the step of sealing the sample
2 within the removable reaction chamber.

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1 32.19 The method according to claim 30 wherein the removable reaction chamber includes
2 an optical interface through which the excitation beam is transmitted from the lens to the
3 sample and an air gap separating the optical interface from the sample, the method further
4 including the step of heating the optical interface to prevent condensation of the sample on
5 the optical interface.

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1 33. The method according to claim 24 wherein the nucleic acid amplification reaction is a
2 polymerase chain reaction.

1 34. The method according to claim 24 wherein the nucleic acid amplification reaction is a
2 ligase chain reaction.

1 35. The method according to claim 24 wherein the nucleic acid amplification reaction is a
2 polymerase chain reaction and wherein the first and second fluorescent indicators are
3 covalently attached to an oligonucleotide having a nucleotide sequence complementary to a

portion of a strand of the amplification reaction product, the second fluorescent indicator quenching the fluorescence of the first fluorescent indicator.

36. A method for monitoring the formation of nucleic acid amplification reaction products in a plurality of samples in real time comprising:
adding samples containing a nucleic acid sequence to be amplified to a plurality of sample holders;
transmitting excitation beams into the plurality of sample holders which illuminate a volume of each sample, each sample including a first fluorescent indicator which produces a first fluorescent signal when illuminated by the excitation beam whose intensity is proportional to the concentration of amplification reaction product in the sample and the volume of the sample illuminated by the excitation beam, and a second fluorescent indicator homogeneously distributed throughout the sample which produces a second fluorescent signal when illuminated by the excitation beam whose intensity is proportional to the volume of the sample illuminated by the excitation beam; and
measuring the intensities of the first and second fluorescent signals of each of the samples.

37. The method according to claim 36 wherein at least two different first fluorescent indicators having different first fluorescent signals are used amongst the plurality of samples, the step of measuring the intensity of the first fluorescent signal including measuring the different first fluorescent signals of the at least two different first fluorescent indicators.

38. A method for monitoring the formation of a plurality of nucleic acid amplification reaction products in a sample in real time comprising:
adding to a sample holder a sample containing a plurality of different nucleic acid sequences to be amplified,
transmitting an excitation beam into the sample which illuminates a volume of the sample, the sample including a plurality of first fluorescent indicators which each produce a

7 first fluorescent signal when illuminated by the excitation beam whose intensity is
8 proportional to the concentration of a particular amplification reaction product in the sample
9 and the volume of the sample illuminated by the excitation beam, and a second fluorescent
10 indicator homogeneously distributed throughout the sample which produces a second
11 fluorescent signal when illuminated by the excitation beam whose intensity is proportional to
12 the volume of the sample illuminated by the excitation beam; and
13 measuring the intensities of the first and second fluorescent signals. --.